On-line monitoring of fermentation of 2G bioethanol

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ON-LINE MONITORING FERMENTATION OF 2G BIOETHANOL

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INTRODUCTION. For many years, cellulosic-ethanol has been considered as an alternative to fossil and non-cellulosic fuels. However, the operational challenges and the lack of understanding of this process often results in poorly operated fermentations that limit their potential to compete with non-cellulosic fuels.

OBJECTIVE. This project aims at implementing spectroscopic-based real-time monitoring methods and to combine them with mechanistic models to predict the dynamics of cellulose-to-ethanol fermentation and to early detect possible contaminations by lactic acid bacteria, achieving an operation closer to the optimal conditions.

FERMENTATION STEPS IN 2G BIOETHANOL

BATCH PHASE
Detoxification phase and cell growth.

FED-BATCH PHASE
Anaerobic phase, to produce of ethanol.

BATCH PHASE
Anaerobic phase until depletion of substrates.

During the initial batch phase, the cell culture is adapted to the medium and detoxifies the inhibitors. Once it starts growing, a fed-batch phase starts and ethanol is produced. The feeding rate is adjusted so it does not exceed the detoxification capacity of the cell culture. Finally, a batch phase runs until the glucose and xylose are finished.

CHALLENGES OF 2G BIOETHANOL FERMENTATION

SUBSTRATE VARIABILITY
The substrate changes its composition from due to different cultivars and weather conditions.

INHIBITORS
The inhibitors produced during the pretreatment inhibit the yeast and extend the length of the lag phase.

CONTAMINATIONS
The fermentation is non-sterile increasing risk of contamination by lactic acid bacteria (LAB).

OXYGEN
Presence of oxygen during the fed-batch phase promotes the production of biomass and reduces the EtOH yield.

MONITORING OBJECTIVES

1. Fermentation dynamics
Due to the substrate variability, the dynamics and end point of the fermentation change from batch to batch.

2. Concentration of inhibitors
Knowing the concentration of inhibitors is used to adjust the feed rate during the fed-batch phase to avoid concentrations of inhibitors that exceed the detoxification capabilities of the cell culture.

3. Early detection of LAB
Early detection of contaminations is fundamental to avoid consuming the substrate to produce LAB and lactic acid instead of ethanol.

4. Presence of oxygen
It is usually not a problem, but it is important to detect because it directs the carbon flow towards biomass instead of ethanol.

CURRENT MONITORING APPROACHES

pH. Monitoring and controlling pH is used to lower the inhibitory effects of the weak acids. However, alone, pH is difficult to correlate with the dynamics of the fermentation, with the concentration of non-acidic inhibitors (i.e. furfural or HMF), and with the LAB concentration.

Off-gas. CO_2, O_2 and ethanol are monitored in the off-gas. CO_2 and ethanol can be correlated with the dynamics of the fermentation but cannot be used to measure the concentration of inhibitors or to detect contaminations

SPECTROSCOPY AS A MONITORING METHOD

The current monitoring methods do not match almost any of the previous monitoring objectives resulting in poorly operated fermentations.

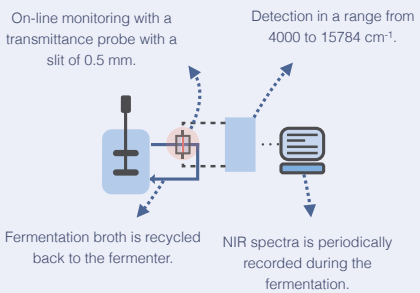
To run the fermentation close to the optimal conditions, it is necessary to add monitoring methods that can deliver information regarding the dynamics of the process in real time.

Vibrational spectroscopy (NIR, MIR) and Raman spectroscopy can detect several key compounds simultaneously, and combined with the current methods, can deliver a good description of the dynamics of the information.

IMPROVING THE REAL TIME MONITORING OF CELLULOSE TO ETHANOL FERMENTATIONS

STEP 1. ON-LINE MONITORING OF KEY COMPOUNDS USING SPECTROSCOPY

EXPERIMENTAL SET UP FOR ON-LINE MONITORING



PLS models are developed to predict the concentrations of glucose, xylose, ethanol.

EXPERIMENTAL DESIGN.

STEP 2. DEVELOPMENT OF A MATHEMATIC MODEL

Development and validation of a mathematical model describing the growth of yeast, the interactions with the different inhibitors and a simplified model of the growth of LAB.

CONCEPTUALIZATION OF THE MECHANISTIC MODEL

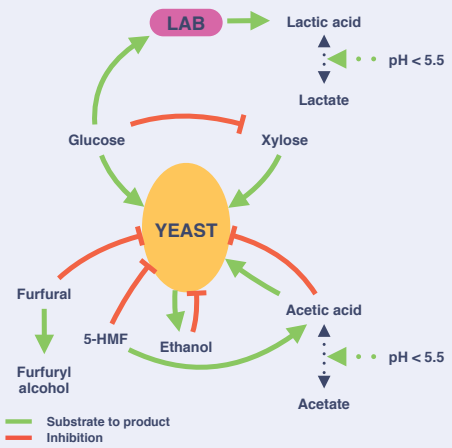
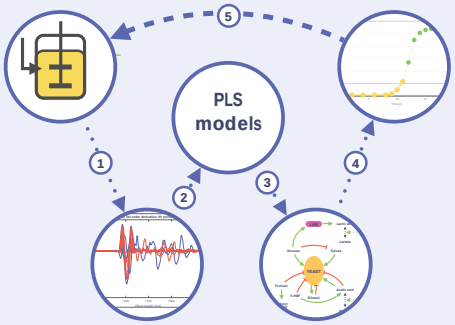


Figure adapted from the model developed by Mauricio-Iglesias, Miguel *et. al. (2015).*

STEP 3. COMBINING THE ON-LINE MEASUREMENTS WITH THE MECHANISTIC MODELS.



1. Real time spectroscopic measurements.

2. The spectra are analyzed using PLS models and the concentrations of key metabolites are predicted.

3. The predicted concentrations are used as input for the mechanistic model.

4. Non-measurable state variables are calculated using the model.

5. Based on the predicted results, a control action can be taken to optimize each fermentation.

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